

LEASE
ENTER
88
7/15/04

AMENDMENTS TO THE CLAIMS

This listing of claims will replace all prior versions and listings of claims in the application:

LISTING OF CLAIMS:

1.-9. (canceled).

10. (previously presented) A method for determining whether a selected DNA molecule encodes a gene expression region, said method comprising:

(a) obtaining RNA transcripts from an organism which comprises said DNA molecule, and

(b) screening said RNA transcripts for an RNA transcript that is complementary to said selected DNA molecule, wherein the nucleotide sequence of said selected DNA molecule is known, to thereby determine whether said selected DNA molecule encodes a gene expression region.

11. (previously presented) The method according to claim 10, wherein said selected DNA molecule consists of from 100 to 200 nucleotides.

12. (previously presented) The method according to claim 10, wherein said screening comprises:

(a) amplifying the RNA transcripts using a first oligonucleotide primer and a second oligonucleotide primer, wherein said first primer is complementary to a sequence of at least 10 continuous nucleotides located at or near the 3'-end of said selected DNA molecule, and said

second primer is homologous to a sequence of at least 10 continuous nucleotides located at or near the 5'-end of said selected DNA molecule, and

(b) detecting an amplification product of (a) complementary to said selected DNA molecule, to thereby screen said RNA transcripts for an RNA transcript that corresponds to said selected DNA molecule.

13. (previously presented) The method according to claim 12, wherein said amplifying comprises:

(a) forming a RNA-DNA duplex comprising one of said RNA transcripts and a complementary DNA molecule adhered thereto, said duplex is formed by synthesizing a first DNA molecule complementary to at least a portion of one of said RNA transcripts using (1) said first oligonucleotide primer to prime synthesis of said first DNA molecule, (2) RNA-dependent DNA polymerase and (3) one of said RNA transcripts as a template, to thereby form an RNA-DNA duplex,

(b) preparing a single-stranded DNA molecule from said RNA-DNA duplex of (a) by hydrolyzing the RNA transcript of said RNA-DNA duplex using ribonuclease H,

(c) forming a doubled-stranded DNA molecule comprising the single-stranded DNA molecule of (b) and a complementary DNA molecule thereto, said doubled-stranded DNA molecule is formed by synthesizing a second DNA molecule complementary to at least a part of said single-stranded DNA molecule of (b) using (1) said second oligonucleotide primer to prime the synthesis of said second DNA molecule, wherein said second primer further comprises an RNA-transcribable promoter sequence at its 5'-end, (2) DNA-dependent DNA polymerase, and

(3) the single-stranded DNA molecule of (b) as a template, to thereby form a double-stranded DNA molecule,

(d) forming an RNA transcription product from said double-stranded DNA molecule of (c) using RNA polymerase, wherein RNA transcription is primed from the RNA-transcribable promoter sequence, and

(e) repeating steps (a) to (d) using said RNA transcription product of (d) as a template for the formation of the RNA-DNA duplex of (a).

14. (previously presented) The method according to claim 12, wherein said method comprises:

(a) performing said amplifying in a reaction solution comprising an oligonucleotide probe that specifically binds to said amplification product, wherein said probe is labeled with an intercalating fluorescence dye and said probe consists of a sequence that is not complementary to either the first or second oligonucleotide primer, and

(b) performing said detecting by measuring a change in a fluorescence characteristic of the reaction solution after the amplification.

15. (previously presented) The method according to claim 14, wherein said probe is complementary to at least a portion of the sequence of said amplification product, and wherein said probe is labeled with an intercalating fluorescence dye that has a differential fluorescence characteristic depending on whether said probe exists in an unbound single-stranded state or in a bound duplex with said amplification product.

16. (currently amended) A method for determining which portions of a selected DNA molecule encode a gene expression region, said method comprising repeatedly performing the method of claim 10 ~~claim 1~~ on successive DNA fragments of said selected DNA molecule.